Delegate	Title	Author(s)	Talk Abstract
Claudio Alonso	The impact of microRNA regulation on neural development and behaviour	Claudio R. Alonso University of Sussex	The cellular components of the nervous system form under the directions of the genes. This suggests that the systematic removal of genes might reveal some of the principles underlying the development of neural systems and how these produce behaviour. We have taken this 'genetic' approach to study the roles played by small non-coding RNAs (microRNAs) during the formation of the Drosophila nervous system. We found that mutation of a single microRNA locus can have specific behavioural effects and affect the ability of Drosophila larvae to correct their orientation if turned upside down ("self-righting"). A key microRNA target involved in this behaviour is the Hox gene Ultrabithorax, whose derepression in two metameric motorneurons (SR-MNs) leads to self-righting defects. Neural activity analyses revealed that these motorneurons have different neural activity patterns in wild type and miRNA mutants, and artificial manipulation of SR-MNs activity results in changes in self-righting behaviour. Conditional expression and cellular analysis experiments suggest, unexpectedly, that the effects of miRNA regulation are primarily behavioural, i.e. concerning the workings of the network rather than underlying its formation. Furthermore, several genome-wide genetic screens reveal that miRNA regulation has pervasive effects on SR as well as on other early motor behaviours suggesting that miRNAs play multiple roles in behavioural control. I will discuss the implications of our findings for the understanding of the genetic basis of behaviour.
Jonas Bittern	The influence of glia on larval locomotion	Jonas Bittern[1], Nils Otto[2], Sören Klemm[3], Benjamin Risse[3], Xiaoyi Jiang[3], Christian Klämbt[1] [1] Institute for Neurobiology, University of Muenster	The nervous system is a highly complex structure built by two cell types, neurons and glia and their interaction is essential for proper brain function. Glial cells provide trophic support for neurons and participate in neurotransmitter homeostasis at the synapse.

		[2] Centre for Neural Circuits and Behavior, University of Oxford [3] Department of Computer Science, University of Muenster	Furthermore, they are thought to actively modulate neuronal activity via so called gliotransmitters. The underlying molecular mechanisms, however, remain largely unknown. We use Drosophila larval locomotion as readout for neural function. High-throughput approaches, however, require automated data analysis. This is facilitated by a frustrated total internal reflection (FTIR)-based imaging method (FIM) to image larvae with a high signal-to-noise ratio to obtain unprecedented high contrast images we established in our lab. Using FIM we screened for glial genes required in the Drosophila CNS for normal locomotor behavior. Here we found the mitochondrial sulfite oxidase Shopper affecting glutamate homeostasis in ensheathing glia which then acts on neuronal network function. In order to better screen for modulatory functions of glia we have developed two alternative locomotion based screening paradigms: Thermogenetic activation of the Goro circuit (Ohyama et al., 2015) triggers a stereotypic rolling behavior in Drosophila larvae which can easily be visualized using FIM. This rolling behavior is quantified upon glial knockdown of single genes to provide a hint for the requirement of the respective glial gene for proper transmission of the neuronal signal. Feeding behavior is controlled by circuit that integrates information on internal and external states. To determine how glia affects the activity of this neuronal circuit, we measure larval food intake in an online fashion using FIM2c.
Alastair Garner	Interneuron populations coordinating locomotor behaviour in D. melanogaster larvae	Alastair Garner*, Jiayi Zhu, Yassine Rahmouni, Tomoko Ohyama	Neural circuit motifs for locomotor behaviours are highly conserved across species. These circuits are hardwired to encode the coordination of behaviour, which encompasses sensory integration, selective motor pool activation and intrinsic motor sequencing. Previous research has revealed circuit mechanisms for motor timing (notably central pattern generators), but less is known about the mechanisms underlying the recruitment of distinct motor ensembles. Recent studies have highlighted the larval fruit fly (Drosophila melanogaster) as a powerful model for studying

			locomotion, as critical circuit nodes sufficient for initiating a stereotyped escape behaviour repertoire (bending, rolling and fast-crawling) have already been described. Using this model, we aim to dissect the components of the interneuron circuitry that coordinates escape behaviours. To address this issue, we use optogenetic manipulations in vivo to interrogate the contribution of ventral nerve cord interneurons to behavioural performance. We report the discovery of two lineage-related premotor neuron populations that affect the expression of escape behaviours. Perturbing the function of these neurons induces abnormal behaviour sequencing and suppression of specific components of the behavioural repertoire. We conduct additional behavioural experiments and morphological analysis to further characterise these populations. Our preliminary data suggest that distinct interneuron populations modulate the activity of distinct motor pools during locomotion.
Carlo Giachello	Nitric Oxide participates in motor network tuning during an embryonic critical period	Carlo N. G. Giachello, Yuen Ngan Fan and Richard A. Baines Faculty of Biology, Medicine & Health, University of Manchester, Manchester, M13 9PT, UK	Neural circuits are most sensitive to activity-dependent tuning during specific time windows, often termed critical periods. We previously identified a critical period in the development of the Drosophila larval motor circuit using optogenetics. Whole-cell recordings from identified aCC motoneurons showed that neural activity manipulation during late embryogenesis affects post-embryonic neural network stability, evidenced by a significant increase in duration of excitatory synaptic input currents. A behavioural analysis revealed that embryonically-manipulated third instar larvae recover slowly after electrocution, thus suggesting a strong correlation between excitatory synaptic inputs and the ability to recover from electroshock. However, the physiological mechanisms involved in this process remain unknown. We have identified Nitric oxide (NO), and its canonical signalling pathway, to be a key part of the activity-induced changes observed during the critical period. Embryonic exposure to NO inhibitors, prior to optogenetic treatment, is sufficient to abolish the

			destabilising effect of activity manipulation. Moreover, we found that the selective potentiation of NO levels in motoneurons, during embryogenesis, is sufficient to recapitulate the same features induced by optogenetic manipulation, suggesting a crosstalk between motoneurons and their synaptic partners. The characterisation of the larval locomotor circuit is still under investigation. Recently, a cholinergic pre-motor interneuron termed A27h has been found to monosynaptically excite aCC. We show that the synaptic strength of the A27h>aCC connection is drastically impaired by embryonic activity-manipulation during the critical period. At the same time, the intrinsic excitability of A27h and aCC neurons seems to be oppositely modulated affecting their ability to fire action potentials. Similar changes to both neurons can be produced by perturbing NO levels during embryogenesis. Taken together, our findings show that the NO pathway is involved in neural network tuning during a critical period in embryogenesis by functionally adjusting both synaptic connectivity and membrane excitability between synaptic partners.
Kristina Klein	Neural circuits of classical and operant conditioning	Kristina Klein[1, 2], Elise Croteau-Chonka[1, 2], Jean-Baptiste Masson[1, 3], Marta Zlatic[1, 2] [1] Janelia Research Campus, Howard Hughes Medical Institute, Ashburn, VA, USA [2] Department of Zoology, University of Cambridge, Cambridge, United Kingdom [3] Decision and Bayesian Computation, Pasteur Institute, Paris, France	Animals have to adapt to a changing environment to improve their chances of survival. Associative learning is the process in which an animal learns to predict an unconditioned stimulus, for example a punishing or rewarding event, by the occurrence of a conditioned stimulus. Classical conditioning, where the conditioned stimulus takes the form of a sensory stimulus such as a visual cue or an odor, is relatively well-understood, and in Drosophila memory formation has been linked to the mushroom body. By contrast, operant conditioning is the process by which an animal learns to associate its own behavior with punishment or reward, leading to suppression or enhancement of certain actions in the future. The neural mechanisms underlying operant conditioning are much less clear, and it has remained an open question whether Drosophila larvae are capable of operant learning. To identify neurons signalling positive or negative valence in a learning context, we have

			performed an optogenetic olfactory conditioning screen. We show that pairing of an odor with activation of different subsets of serotonergic neurons is sufficient to induce both appetitive and aversive classical conditioning. Using a closed-loop tracker with online behavior detection and optogenetic LEDs, we have tested candidate valence-conveying neurons identified in the olfactory conditioning screen for their potential to serve as an unconditioned stimulus for operant learning, and provide examples for both operant reward conditioning and operant punishment conditioning of bend direction in the larva.
Mason Klein	Dissecting the exploration and navigation strategies of individual larvae over short and long time scales	Mason Klein (Assistant Professor, University of Miami)	How the internal properties of animals combine with their response to external environmental stimuli is an important question in understanding how the brain operates. In Drosophila larva crawling, the animals exhibit exploratory behavior as they search for food, while at the same time responding to environmental stimuli. Or, for example, while crawling on a stimulus gradient the inherent bias of an individual larva to turn or drift predominantly leftward or rightward ("handedness") would combine with stimulus information to produce the overall behavioral response. We examine these cases by measuring 2D navigation in larvae. Their behavior can be classified as "diffusion," even along the axis of strong navigation, and their long term behavior is consistent with a Markov state model. Using handedness as a measure of an individual animal's internal bias, we find that there is significant bias in individual turn and drift direction, but that turn and drift handedness are uncorrelated. This lack of correlation explains why even strongly left- or right-turning larvae on average have similar diffusion rates as unbiased turners. Both handednesses are weakly persistent, even across instars. Finally, we show that the internal bias (handedness) strongly affects individual turning decisions in the presence of a temperature gradient, suggesting that inherent traits of individuals are needed for a more complete understanding of navigation. To uncover these results we have employed several

			novel methods: (1) color-tagging individual larvae with dyed food to measure their behavior for 5-6 days while maintaining identity; (2) running Monte Carlo simulations that include drift in between sharp turns, by stitching together track segments drawn from a large (~20,000) empirical collection; (3) using a customized robot that can pick up and place larvae around an arena, which greatly increases the information gathered from individual larvae, and should be readily applicable to many other larva experiments.
Hiroshi Kohsaka	A modular structure in premotor circuits for bidirectional axial locomotion	Hiroshi Kohsaka 1, Maarten F. Zwart 2, 4, Akira Fushiki 2, 5, Richard D. Fetter 2, James W. Truman 2, 6, Albert Cardona 2 and Akinao Nose1,3 1 Department of Complexity Science and Engineering, Graduate School of Frontier Science, the University of Tokyo, 5-1-5 Kashiwanoha, Kashiwa-shi, Chiba-ken, 277-8561, Japan 2 HHMI Janelia Research Campus, Ashburn, VA 20147, USA 3 Department of Physics, Graduate School of Science, the University of Tokyo, 7-3-1 Hongo, Bunkyo-ku, Tokyo, 133-0033, Japan 4 School of Psychology and Neuroscience, University of St Andrews, KY16 9JP Scotland, United Kingdom 5 Departments of Neuroscience and Neurology, Zuckerman Mind Brain Behavior Institute, Columbia University, New York, NY., USA. 6 Friday Harbor Laboratories, University of Washington, Friday Harbor, WA. 98250, USA	Animal locomotion requires spatiotemporally coordinated contraction of muscles throughout the body. Here, we investigate how contractions of antagonistic groups of muscles are intersegmentally coordinated during bidirectional crawling of Drosophila larvae. We identify two pairs of higher-order premotor excitatory interneurons that are present in each abdominal neuromere and intersegmentally provide feedback to the adjacent neuromere during motor propagation. The two feedback neuron pairs are differentially active during either forward or backward locomotion but commonly target a group of premotor interneurons that together provide excitatory inputs to transverse muscles and inhibitory inputs to the antagonistic longitudinal muscles. Inhibition of either feedback neuron pair compromises contraction of transverse muscles in a direction-specific manner. Our results suggest that the intersegmental feedback neurons coordinate contraction of synergistic muscles by acting as delay lines representing the phase lag between segments. The identified circuit architecture also shows how bidirectional motor networks can be economically embedded in the nervous system.
Kai Li	Elucidation of neural circuit mechanism integrating noxious stimulus and ambient	Kai Li, Akira Murakami, Tadashi Uemura and Tadao Usui	Avoidance of harmful stimuli is essential for animals' survival. Such a nociceptive response is a protective mechanism to escape from damage to the tissue. We have been interested in Class IV dendritic arborization neurons (C4da neurons) in Drosophila

	temperature sensation in Drosophila	Uemura Lab., Graduate School of Biostudies, Kyoto University	larvae, which are polymodal nociceptors responsible for thermal, mechanical and light sensation (Chin and Tracey, Curr. Biol., 2017; Onodera et al., eLife, 2017; Terada et al., eLife, 2016). The activation of these neurons leads to distinct behaviors, depending on the inputs. For example, the noxious thermal stimulation triggers a stereotyped avoidance response named rolling escape behavior. However, the downstream neural processing mechanism of harmful stimuli is still elusive. Interestingly, a group of thermosensory neurons in the larval brain can also induce the rolling behavior, which indicates that there is a central input for the behavior (Luo et al., Nature Neurosci., 2016). Why does larva have such thermosensors deep in the brain? How is this central thermo-sensation transduced to the downstream? How does such descending input from brain affect the peripheral nociceptive sensation? How are two input integrated by the downstream circuits and contribute to the behavioral decision? Most likely, such integration of multiple inputs happens in the ventral nerve cord and may confer certain benefits to the organismal survival in natural environments. Our behavioral tests and imaging results suggested that these central thermosensors might modulate synaptic transmission from C4da neurons to their downstream neurons and affect the function of the larval nociceptive circuit. Keywords: Drosophila, nociception, thermo-sensation, avoidance behavior, neural circuit, ventral nerve cord
Matthieu Louis	Sensorimotor control of reorientation behavior during larval chemotaxis: running or stopping	Matthieu Louis, University of California Santa Barbara	Larval chemotaxis consists of an alternation between runs and reorientation maneuvers. The detection of positive gradients in the concentration of an attractive odor prolongs running while negative gradients promote stopping and turning. In a behavioral screen, we identified an olfactory descending neuron (PDM-DN) that plays an essential role in the sensorimotor conversion of dynamic olfactory

			inputs into the release of stops and turns. Using electron microscopy and functional imaging, we mapped the main pathway that connects the PDM-DN neuron to the peripheral olfactory sensory neurons down to pre-motor circuits in the ventral nerve cord. Our functional analysis clarifies the neural-circuit computation that transforms graded sensory input into action selection to perform navigation.
Dennis Pauls	Reward signaling in a recurrent circuit of dopaminergic neurons and Kenyon cells in the Drosophila larva	Radostina Lyutova, Maximiilan Pfeuffer, Dennis Segebarth, Jens Habenstein, Astrid Rohwedder, Mareike Selcho, Christian Wegener, Andreas Thum, Dennis Pauls	Dopaminergic neurons in the brain of the Drosophila larva play a key role in mediating reward information to the mushroom bodies during appetitive olfactory learning and memory. Using optogenetic activation of Kenyon cells we provide evidence that a functional recurrent signaling loop exists between Kenyon cells and dopaminergic neurons of the primary protocerebral anterior (pPAM) cluster. An optogenetic activation of Kenyon cells paired with an odor is sufficient to induce appetitive memory, while a simultaneous impairment of the dopaminergic pPAM neurons abolishes memory expression. Thus, dopaminergic pPAM neurons mediate reward information to the Kenyon cells, but in turn receive feedback from Kenyon cells. Further, our data suggests that feedback signaling is dependent on short neuropeptide F (sNPF), the only neuropeptide known - so far - to be expressed in Kenyon cells. Finally, we show that an artificial activation of the mushroom body circuitry during training increases the persistence of an odor-sugar memory.
Nino Mancini	Function of the anterior paired lateral (APL) neuron in associative olfactory learning in larval Drosophila	Nino Mancini, Michael Schleyer, Bertram Gerber	Inhibitory systems are important controllers of sensory systems and behaviour, allowing the processing of relevant information against environmental noise, and the selection of adaptive motor actions from a pool of competing behavioural options. Several studies have shown the critical role of GABAergic synaptic inhibition in odour processing, olfactory learning and behavior in invertebrates, including Drosophila melanogaster. In this project, we focus on a single, GABAergic anterior paired lateral (APL) neuron, identified in both adult and larval Drosophila.

			Although the role of APL in memory acquisition and retrieval has been investigated in adults, the lack of a defined circuitry limits the interpretation of behavioural and physiological data. Larval Drosophila, however, offers such possibilities because of its simple olfactory system that is well characterized at synaptic resolution and without cellular redundancy. Using a combination of behavioural analysis, optogenetics and connectomics, we aim to understand how APL inhibitory processes are organized in the larval brain and how they modulate associative olfactory memory formation and retrieval. We discovered, surprisingly, that activating APL optogenetically is sufficient to establish a reward memory. We follow up on this asking whether this rewarding effect requires intact GABA synthesis in APL and working in collaboration with colleagues from Würzburg University and the NIH contributing to physiological expertise.
Mirna Mihovilovic	Recording neural activity in unrestrained animals with 3D tracking two photon microscopy	Mirna Mihovilovic	Optical recordings of neural activity in behaving animals can reveal the neural correlates of decision-making, but such recordings are compromised by brain motion that often accompanies behavior. Two-photon point scanning microscopy is especially sensitive to motion artifacts, and to date, two-photon recording of activity has required rigid mechanical coupling between the brain and microscope. To overcome these difficulties, we developed a two-photon tracking microscope with extremely low latency (360 µs) feedback implemented in hardware. We maintained continuous focus on neurons moving with velocities of 3 mm/s and accelerations of 1 m/s both in-plane and axially, allowing high-bandwidth measurements with modest excitation power. We recorded from motor- and inter- neurons in unrestrained freely behaving fruit fly larvae, correlating neural activity with stimulus presentation and behavioral outputs, and we measured the light-induced depolarization of a visual interneuron in a moving animal using a genetically encoded voltage indicator. Our technique can be extended to stabilize recordings in a variety of moving substrates.

Birgit Michels	Memory enhancement by ferulic acid ester across species	Birgit Michels[1], Hanna Zwaka[1,2], Ruth Bartels[2], Oleh Lushchak[3], Katrin Franke[4], Thomas Endres[5], Markus Fendt[6,11], Inseon Song[7], May Bakr[7], Tuvshinjargal Budragchaa[4], Bernhard Westermann[4], Dushyant Mishra[8], Claire Eschbach[8], Stefanie Schreyer[2], Annika Lingnau[2], Caroline Vahl[2], Marike Hilker[2], Randolf Menzel[2], Thilo Kähne[9], Volkmar Leßmann[5,11], Alexander Dityatev[5,7,11], Ludger Wessjohann[4], and Bertram Gerber[1,10,11] 1 Leibniz Institute for Neurobiology (LIN), Department Genetics of Learning and Memory, Magdeburg, Germany. 2 Free University Berlin, Institute of Neurobiology, Berlin, Germany. 3 Precarpathian National University, Department of Biochemistry, Ivano-Frankivsk, Ukraine. 4 Leibniz Institute of Plant Biochemistry (IPB), Department of Bioorganic Chemistry, Halle/ (Saale), Germany. 5 Otto von Guericke University, Medical Faculty, Institute of Physiology, Magdeburg, Germany 6 Otto von Guericke University, Medical Faculty, Institute for Pharmacology and Toxicology, Magdeburg, Germany. 7 German Center for Neurodegenerative Diseases (DZNE), Molecular Neuroplasticity Group, Magdeburg, Germany. 8 University of Würzburg, Biocenter Am Hubland, Department of Genetics and Neurobiology, Würzburg, Germany.	Cognitive impairments can be disturbing if not devastating for the quality of life, and any means of preventing or counteracting them is of value. We exploit the potential of the plant Rhodiola rosea that is used to this end in folk medicine and identify the constituent ferulic acid eicosyl ester (FAE-20) as a memory enhancer. Food supplementation with dried root material from Rhodiola rosea dose-dependently improves associative memory scores in larval Drosophila and prevents age-related memory decline in adult flies. Task-relevant sensory-motor faculties remain unaltered. Using a combined bioassay-guided fractionation and bioinformatics approach, we identify Rhodiola-derived FAE-20 as a candidate compound. Indeed, de novo synthesized FAE-20 is effective as a memory enhancer in both Drosophila larvae and in aged adult flies, and can counteract genetically-induced early-onset loss of memory function in young flies. Furthermore, treatment with FAE-20 increases excitability in mouse hippocampal CA1 neurons and leads to more stable memory upon contextual fear conditioning. Given the conserved effects of FAE-20 from maggots to mouse, and given the utility of Rhodiola as the FAE-20 source-plant in traditional human medicine, these results hold potential for clinical application.

		9 Otto von Guericke University, Institute of Experimental Internal Medicine, Magdeburg, Germany. 10 Otto von Guericke University, Institute of Biology, Magdeburg, Germany. 11 Center for Behavioral Brain Sciences (CBBS), Otto von Guericke University, Magdeburg, Germany	
Liria Masuda-Nakagawa	Octopamine regulates behavioral odor discrimination in Drosophila larva	Alex D McLachlan[1], Marcella Montagnese[1], J Y Hilary Wong[1], Bo Angela Wan[1], Liria M Masuda-Nakagawa[1]	Insect mushroom bodies (MBs) are higher brain centers essential for associative olfactory learning. The calyx (input region) of the MBs in third instar larval Drosophila, is organised in around 34 calyx glomeruli, each receiving stereotypic innervation of a single olfactory projection neuron (PN). MB neurons, KCs, are combinatorial integrators of these multiple inputs. We previously showed that the selectivity of odour representation might be regulated by an inhibitory feedback neuron, the larval APL, the sole detectable GABAergic input in the larval calyx, and now aim to understand how other calyx extrinsic neurons integrate with this circuitry. A second set of neurons innervating the calyx comprises two octopaminergic (OA) neurons, the sVUMmd1 and sVUMmx1 neurons, which originate in the mandibular and maxillary segments respectively, in the subesophageal zone (SEZ). Their calyx terminals appeared presynaptic, and multicolor flipout showed that they both innervate the calyx widely. GRASP experiments suggested that they synapse on KCs, PNs and the APL, and with another class of calyx output neurons characterized by odd expression. To test which neurons receive OA input, we analyzed localization of endogenous GFP-tagged OA receptors. We found that OAMB is localized in PN terminals, suggesting that OA may influence calyx function via PN activity. We therefore imaged the effects of optogenetically activating Tdc2 OA neurons on odor-induced responses in PN termini , and found potentiation of PN responses.

			We have used optogenetic manipulation of a small OA neuron subset that includes the sVUM1s, and tested the consequences for discrimination behavior. Activation of these neurons impaired discrimination between a pair of similar odours in olfactory choice learning, without impairing underlying learning ability. Therefore, octopamine released by VUM1 neurons in the calyx appears to regulate the sensitivity of the calyx to sensory input.
Larisa Neagu-Maier	Taste coding principles in Drosophila larva	G. Larisa Neagu-Maier[1], Felix Meyenhofer[1], Wanze Chen[2], Marjan Biocanin[2], Johannes Bues[2], Bart Deplancke[2], Simon G. Sprecher[1] [1]Department of Biology, University of Fribourg, 1700 Fribourg, Switzerland [2]Laboratory of Systems Biology and Genetics, Institute of Bioengineering (IBI), School of Life Sciences, EPFL and Swiss Institute of Bioinformatics (SIB), 1015 Lausanne, Switzerland	Deciding if the food is good or bad to eat is a matter of survival for most animals. Hence, the ability to discriminate between different tastes is innate - we are born to be attracted to sweet and savory things and to be averse to bitter and sour aliments, as sweet taste can indicate a rich-nutrient food source while bitter can signal the presence of noxious food. The objectives set out within our study are first steps towards shaping up a complete map with spatial, physiological and molecular information for the Drosophila larval primary taste organ. The restricted number of taste neurons in the Drosophila larva makes the dissection of taste encoding principles a conceivable endeavor. We are generating a complete functional 3D map of the main gustatory organ in the larva (termed terminal organ, TO) comprising physiological responses and receptor expression. Using a custom-made microfluidic chip that allows in-vivo functional imaging of the taste neurons while stimulating the larva with different tastants we are reconstructing the physiological response profiles at cellular resolution (van Giesen, Neagu-Maier et al., 2016b). Additionally, we use single cell transcriptomics by implementing a modified Dropseq method adapted for low number of cells to decipher the molecular fingerprint of individual receptor neurons, providing a sensory receptor gene expression map.

			Our investigation further highlights the multimodal tuning of larval primary taste neurons and the large co-expression repertoire of sensory receptor genes in this system.
Nick Polizos	The use of temperature to facilitate associative learning and memory retention in Drosophila larvae	Nikolaos T. Polizos Klein Lab: University of Miami	Organisms have evolved the ability to detect various stimuli to successfully navigate the world in which they live. Not all forms of stimuli are equally salient after processing, forming a hierarchy of sensory stimuli. The most salient sensory experiences can be stored and referenced in the form of memory. Memory is particularly interesting in that it allows for behavioral flexibility in direct response to the environment. By examining this interface between an organism and its environment, the effects of natural selection on learning and memory can be better understood. This approach requires a strong background knowledge of the sensory inputs an organism relies upon. The Drosophila melanogaster larval model is ideal for this approach because it has simple body plan yet exhibits a number of easily quantifiable navigational behaviors. Additionally, the mechanisms responsible for larval olfaction, thermotaxis, and phototaxis are being actively researched. In this project we hope to test the associative memory of larva when conditioned with each of these stimuli. By assessing the performance of wild type and mutant larvae it is possible to determine the saliency of each of these stimuli. This information can aid in understanding how prior experiences shape responses to selection pressures.
Quan Yuan	Temporal control of inhibition generates ON and OFF selectivity in Drosophila larval visual circuit	Bo Qin[1], Tim-Henning Humberg[2]*, Anna Kim[1]*, Hyong Kim[1], Jacob Short[1], Fengqiu Diao[3], Benjamin H. White[3], Simon Sprecher[2] and Quan Yuan[1]# [1]National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, MD, 20892, USA [2]Department of Biology, University of Fribourg, Fribourg, Switzerland	ON and OFF selectivity in visual processing is encoded by parallel pathways that respond to either light increments or decrements. Despite lacking anatomical features to support split channels, Drosophila larvae effectively perform visually-guided behaviors. To understand principles guiding visual computation in this simple circuit, we focus on the physiological properties and behavioral relevance of larval visual interneurons and elucidate their functions in visual processing. We find that the ON vs. OFF discrimination in the larval visual circuit emerges through light-elicited cholinergic

		[3]National Institute of Mental Health, National Institutes of Health, Bethesda, MD, 20892, USA *: equal contributions #: Corresponding author and lead contact: quan.yuan@nih.gov	signaling that activates the cholinergic interneuron (cha-lOLP) and inhibits the glutamatergic interneuron (glu-lOLP). Genetic studies further indicate that the reciprocal inhibition between cholinergic and glutamatergic neurotransmission separates the ON and OFF signals through temporal shifts, the disruption of which strongly impacts both physiological responses of downstream projection neurons and dark-induced pausing behavior. Together, our studies identify cellular and molecular substrates for OFF detection in the larval visual circuit and reveal that temporal control of inhibition functions as an effective strategy in generating ON and OFF selectivity without anatomical segregation.
Nadine Randel	Circuit mechanisms for behavioral choice	Nadine Randel, Chen Wang, Harald Hess, Philipp Keller, Albert Cardona, Marta Zlatic	Behavioral choice is essential for the survival of all animals. Nevertheless the neuronal mechanisms at the level of single neurons and whole brain dynamics are poorly understood. A main obstacle to progress is that the behavioral choice circuits are widely distributed, involving many brain regions. Hence investigation of complete neuronal mechanisms necessitate the study of structural connectivity and neuronal dynamics in the whole nervous system. Recent advances in electron microscopy enable the synaptic resolution connectomes for relatively small nervous systems. Likewise, recent advances in light sheet microscopy support the monitoring of neuronal activity in entire nervous systems. However, an unsolved challenge is the direct combination of functional activity maps and synaptic connectivity maps for the same organism. We have developed a methodology to overcome this difficulty, where we perform whole brain functional imaging with subsequent low resolution EM at the same sample, to identify neurons that have interesting activity patterns. We will apply the approach to studying the neuronal mechanisms of behavioral choice, between one of five possible mutually exclusive actions in Drosophila melanogaster larva, that can occur in response

			to the same stimulus (optogenetic activation of nociceptive neurons). The aim is to identify circuit mechanisms that promote one action while suppressing all competing actions. Preliminary results show, that we can detect neurons, whose activity is correlated (or anticorrelated) with each action and we are currently combining the neuronal activity information with the complete connectome of the distributed circuits. This way we can identify candidate circuit motifs that could promote one action and suppress competing ones, such as recurrent excitation, disinhibition, reciprocal inhibition and others. This is the first time that functional and structural information are combined in the same organism to elucidate a complete neuronal mechanisms, at the example of behavioral choice.
Astrid Rohwedder	Brainbase- a larval Standard Brain online ressource	Astrid Rohwedder[1], Katja Bühler[2], Dorit Merhof[3] and Andreas S. Thum[1] 1 Department of Genetics, University of Leipzig, Leipzig, Germany, 2 VRVis Zentrum für Virtual Reality und Visualisierung Forschungs-GmbH, Vienna, Austria, 3 Institute of Imaging & Computer Vision, RWTH Aachen University, Aachen, Germany	In the recent years, the organization of the larval brain of Drosophila has been intensely studied. From the reconstruction of the connectome of a first instar larval brain in EM to the light-microscopical analysis of thousands of different Gal4 driver lines broadened our knowledge. Ultimately, this data is now being integrated in a newly established standard atlas for the larval brain, a five-part approach that includes the generation of an image registration framework, the generation of a larval standard brain, the segmentation and denomination of identified brain structures, the registration of several thousand Gal4 stocks onto the standard brain, and the organization of the obtained information in a web-based open access database called Brainbase. Additional features of this upcoming database will be information of the immunoreactivity of select cells. Up to now, the light-microscopical part of this database is limited to the third instar larva. However, in the future it will be supplemented with first and second instar larval brains.
Michael Schleyer	Characterization of an optogenetically activated	Michael Schleyer*, Alićе Weiglein, Juliane Thöner, Anne Voigt , Timo Saumweber and Bertram Gerber	A hungry animal may use its previous experience with food-associated stimuli to guide its search for food. Once a food source is found, however, it is adaptive to stop searching and rather exploit

	dopaminergic reward signal	Leibniz Institute for Neurobiology, Brenneckestr 6, Magdeburg, Germany *Corresponding author: michael.schleyer@lin-magdeburg.de	the food source. We study the role of a single, identified dopamine neuron in these processes. In their search for food, Drosophila larvae prefer an odor previously paired with food reward relatively more than an odor that previously was presented unpaired with reward. We show that the larvae track down a reward-associated odor only if there is something to gain, i.e. only if the odor predicts more food than currently present. Moreover, after training with odor and sugar reward larvae specifically search for sugar but not amino acids, and vice versa. That means, larvae establish memories that are specific for sugar vs. amino acid rewards - which allows them to organize their search for food according to their current needs. Using a combinational approach of behavior experiments and optogenetic activation, we currently characterize single dopaminergic central brain neurons for their role in establishing and gating associative memories. For the dopaminergic DAN-i1 neuron we find: (1) its activation is sufficient as internal reward signal, even with only one training trial; (2) the valence of the memory established by DAN-i1 is dependent on the relative timing between odor presentation and DAN-i1 activation; (3) DAN-i1 carries a sugar rather than an amino acid reward signal, and therefore training with DAN-i1 makes larvae specifically search for sugar; (4) the microbehavioral 'footprint' of a DAN-i1-induced memory matches that of a sugar-induced memory; (5) the retrieval of a memory established by DAN-i1 is acutely suppressed by DAN-i1 activation. In summary, this single dopamine neuron carries a sugar-specific internal reward signal that can establish memories of opposite valence depending on the relative timing with the odor, and gates the behavioral expression of the established memory. Our findings

			challenge the notion that dopaminergic neurons always carry a common-currency value signal, and reveal an elegant mechanism to prevent further search once the sought-for item is found.
Andreas Thum	The larval standard brain: The reconstruction of the larval memory center at cellular and synaptic resolution		Brains organize behavior. This involves the integration of present sensory input, past experience, and options for future behavior. The insect mushroom body is a paradigmatic case of a central brain structure bringing about such triadic integration. We use larval Drosophila to systematically study these processes at single-cell and single synapse resolution. We use a bipartite approach including serial section electron microscopy and light microscopy analysis of novel genetic tools to reconstruct every single neuron and all synapses in the entire larval brain (connectome). As a proof of principle, we describe a project focusing on the mushroom body, which consists of about 110 instrinsic Kenyon cells, 24 output neurons, 7 dopaminergic input neurons, 4 octopaminergic input neurons, 5 additional input neurons, and a GABAergic feedback neuron per hemisphere. At the synaptic level, we show further subdivision of the mushroom body into 11 functional subunits, all organized by a conserved connectivity motif defined by individual input, output, and intrinsic neurons. We further aim to integrate the data into a newly established standard atlas for the larval brain via a five-part approach. It includes generation of an image registration framework, generation of a brain template, segmentation and denomination of identified brain structures, registration of several thousand Gal4 and split-Gal4 stocks onto the template, and the organization of the obtained information in a web-based open access database. Taken together this work provides a rich picture to support and enhance future studies on the larval brain on multiple levels.

Naoko Tashima	Appetitive and aversive learning of amino acids in larval Drosophila	Naoko Toshima[1,2], Michael Schleyer[1] and Bertram Gerber[1, 3, 4] Authors' affiliations 1 Leibniz Institute for Neurobiology (LIN), Department Genetics of Learning and Memory, Magdeburg, Germany 2 JSPS Overseas Research Fellow 3 Otto von Guericke University Magdeburg, Germany 4 Center of Behavioral Brain Science (CBBS), Magdeburg, Germany	Although amino acids are important nutrients for Drosophila melanogaster, how flies detect amino acids and how the behavioural response to amino acids are regulated are largely unknown. Previously Toshima & Tanimura (2012) found that adult Drosophila enhance the feeding preference to amino acids when they were deprived of amino acids. Contrary to the adult flies, which can survive without obtaining amino acids, larvae continuously require to ingest protein source for growth. Larval brain consists of relatively small number of neurons, that is ten times fewer than adult brain. Nevertheless, larvae are intelligent enough to exhibit associative learning of odours and taste stimuli. Given that associative learning is related to feeding motivation, it is intriguing to ask whether larvae show reward learning to amino acids. Schleyer et al. (2015) showed that sugar and amino acid induce independent appetitive memories. That is, although fructose and aspartic acid induce similar intensity of appetitive memory, fructose memory is not abolished in the presence of aspartic acid. Similarly, aspartic acid memory is abolished in the presence of aspartic acid, but not in the presence of fructose. We then performed learning experiments for 20 individual amino acids, and found that larvae learn all individual amino acids as reward (Kudow et al. 2017). To see further detail of amino acid learning, here we used an amino acid-mixture as the reinforcer. We also tested genetically modified flies to investigate which neurons contribute to amino acid learning.
James Truman	Metamorphosis: coping with the "two mind - two body" problem	James W. Truman1,2 and Jacquelyn Price2. 1Friday Harbor Laboratories, University of Washington, Friday Harbor WA USA; 2Janelia Research Campus, HHMI, Ashburn, VA USA	Once mature, a neuron is expected to maintain a rather stable identity through the life of an organism. An exception to this stability, though, in seen in insects that undergo complete metamorphosis, such as Drosophila. In such insects, some neurons exhibit two sequential forms, one for the larval stage and a second for the adult, with a remodeling process in between. The adult form and function of such cells likely reflects the cellÕs ÒancestralÓ form and function, while the larval form represents a novel, derived

			state. The origin of the novel, larval form of such neurons, though, is unclear. It may have arisen by truncated development with a neuronÕs intermediate developmental stage being maintained and adapted for use in larval circuitry. Alternatively, the larval form may represent a completely new identity for the cell, which then reverts to its ancestral identity at metamorphosis.Using conditional flip-out techniques, we carried out a large-scale comparison of the fates of different interneuron types within the larval CNS through metamorphosis. Our main focus has been on the input and output neurons of the Mushroom Bodies. We find that both strategies are employed to generate the larval circuitry. In some cases, adult MB input or output cells have a similar role in the larva. In other cases, though, we find that some neurons that function in the adult central complex or optic lobes are integral components of the MB input circuitry in the larva. At metamorphosis, their function is then taken over by neurons that are born during larval life, and these temporary MB-related neurons are restructured to assume their ancestral role in the adult.
Tadio Usui	Belly roll, a member of Ly6/α-Neurotoxin/uPAR protein, regulates the activity of nociceptive circuitry that evokes avoidance behavior	Tadao Usui1, Risa Nishimura1, Shumpei Baba1, Kai Li1, Koun Onodera1, Akira Murakami2, Tadashi Uemura1. 1. Graduate School of Biostudies, Kyoto University 2. Graduate School of Informatics, Kyoto University	Adequate behavioral responses to noxious stimuli are essential for organismal survival. Drosophila larvae show characteristic rolling behavior to escape from the mechanical stimuli caused by parasitoid wasps, the ultraviolet rays in sunlight, or the noxious high temperature (Terada et al., 2016; Onodera et al., 2017). We noticed the fact that the magnitude of rolling behavior of wild-type strains are significantly different and then aimed to understand these diversified behavioral responses through identifying responsible genes. We have quantified the rolling behavior upon high temperature stimulation for 38 representative strains of the Drosophila Genetic Reference Panel; and then found 31 candidate loci by genome-wide association analysis. Secondary functional screen by using RNAi knockdown has revealed that belly roll (bero) gene negatively regulates the rolling behavior. The bero gene encodes a Ly6/uPAR protein and is expressed selectively in

			interneurons of the larval central nervous system. Notably, several neurons expressing bero displayed Ca2+ responses upon activation of nociceptors. Furthermore, the optogenetic activation of bero-expressing neurons evoked typical avoidance behavior. We are currently attempting to elucidate the regulatory mechanism via Bero protein in this circuitry.
Rebecca Vaadia	Functional organization of a Drosophila proprioceptive system for feedback during locomotion	Rebecca Vaadia*1, Wenze Li*2, Aditi Singhania3, Samantha Galindo3, Ya- Ting Lei1, Jennifer K Lee4, Katherine L Lee4, Nathan Carpenter4, Venkatakaushik Voleti2, Elizabeth MC Hillman2,5,6, Wesley B Grueber1,4,6 1Columbia University Medical Center, Department of Neuroscience, New York, NY, 2Columbia University, Laboratory for Functional Optical Imaging, Departments of Biomedical Engineering and Radiology, New York, NY, 3Columbia University Medical Center, Department of Genetics and Development, New York, NY, 4Columbia University Medical Center, Department of Physiology and Cellular Biophysics, New York, NY, 5Columbia University, Kavli Institute for Brain Science, New York, NY, 6Columbia University, Mortimer B. Zuckerman Mind Brain Behavior Institute, New York, NY. *Authors contributed equally	1Columbia University Medical Center, Department of Neuroscience, New York, NY, 2Columbia University, Laboratory for Functional Optical Imaging, Departments of Biomedical Engineering and Radiology, New York, NY, 3Columbia University Medical Center, Department of Genetics and Development, New York, NY, 4Columbia University Medical Center, Department of Physiology and Cellular Biophysics, New York, NY, 5Columbia University, Kavli Institute for Brain Science, New York, NY, 6Columbia University, Mortimer B. Zuckerman Mind Brain Behavior Institute, New York, NY. *Authors contributed equally Proprioceptive sensory neurons provide feedback about body position that is essential for coordinated movement. Understanding how body position is dynamically encoded requires knowledge of proprioceptor activity in freely moving animals. Here we applied high-speed volumetric SCAPE microscopy to simultaneously track the position, physical deformation, and activity of multidendritic proprioceptive neurons in crawling Drosophila larvae. Larval crawling consists of periodic segment contraction and relaxation. A majority of proprioceptors showed sequential onset of activity during segment contraction with one neuron activated by segment extension. Different timing of activity from contraction-sensing proprioceptors was related to distinct dendrite terminal targeting, suggesting that dendrite morphology may be key to timing proprioceptor activity. Such dynamics could endow proprioceptors with distinct roles in monitoring the progression of contraction waves and body movements during other behaviors. How terminal

			territories are shaped to support function is not clear. We examine the mechanisms that target a subset of proprioceptors to specific regions of the body wall. We show that proprioceptors and mechanoreceptors target complementary regions of the body wall. Dendrite-dendrite interactions are not required for targeting of proprioceptor dendrites, rather we provide evidence that substrate-derived cues instruct dendrite targeting. Our results reveal how sensory terminal organization of proprioceptors is linked during development to body wall dynamics and demonstrate that the SCAPE method can be used to characterize neural signaling dynamics in freely behaving organisms.
Michael Winding	Towards a complete connectome of the larval central brain	Michael Winding, Akira Fushiki*, Feng Li*, Laura Herren, Javier Valdes Aleman, Ingrid Andrade, Matthew Berck, Casey Schneider-Mizell, Marta Zlatic**, Albert Cardona** *,**, equal contribution	From moment to moment, the nervous system of any animal must answer one question: 'What to do next?' The computations required to answer this question and perform complex behaviors likely involve many interconnected brain regions. However, due to limited connectivity information, many functional and behavioral studies focus on isolated brain regions or neural circuits to understand behavior. To obtain connectivity data, previous studies have used a 1st instar Drosophila ssTEM volume to reconstruct various brain regions, including sensory and projection neurons, the Antennal Lobe, and the Mushroom Body (associative learning center) with synaptic-resolution. Now, we are using this ssTEM volume to generate a complete wiring diagram of the entire central brain of the Drosophila larva. This will be the first full connectome of an insect brain and the most complex brain reconstructed to date. This data will reveal new neural circuits and interactions between previously identified circuits, which are distributed through the whole brain. Here, we present initial findings, including: 1) the full connectome of the Lateral Horn (innate center), 2) a novel layer of ~300 neurons that integrate inputs from the Lateral Horn and Mushroom Body, and 3) initial connectivity data of all brain neurons. This data has revealed novel and exciting circuit motifs, which might implement winner-take-all computations,

			persistent neuronal activity, and interface with premotor regions. In combination with new full-brain functional imaging techniques (see Nadine Randel's abstract), we are on the verge of a deep understanding of complex, brain-wide circuit computations.
Alice Weiglein	Visualization of a full body Drosophila larva	Aliće Weiglein[1,3], Oliver Kobler[2], Bertram Gerber[1,3,4*]	The Drosophila larva depicts a commonly used model organism to study how a behavioural output is brought about by the brain, especially since it possesses a relatively simple and easily accessible brain which can be examined in its entirety by light and electron microscopy techniques. Indeed, research is almost exclusively focused on the brain since like in the vast majority of life science research does not, and often cannot, transgress the boundaries of a given organ system. Thus, the databases documenting the anatomy of the Drosophila nervous systems feature information on exclusively the nervous system, while the rest of the body is literally thrown away during sample preparation. Similarly restricted are the databases documenting the expression of transgenic driver strains which depict the basis for practically all current research in Drosophila. This can lead research badly astray. To overcome this troubling condition we aim at visualizing a full body intact Drosophila larva. For this approach we use solvent-based clearing methods combined with state-of-the-art light-sheet microscopy that allows us the detection of transgenically expressed fluorescent proteins in the context of the complete larval body. Additionally, we adapted the clearing procedure to use nanotags to improve the signal-to-noise ratio and to be able to use different markers for different cells within the same specimen. Anatomical information about a selected number of driver lines could in the longer run be mapped in a full body standard larva. Author affiliations: *1Leibniz Institute for Neurobiology, Department of Genetics, Magdeburg; 2Leibniz Institute for Neurobiology, Department Special Lab Electron and Laserscanning Microscopy, Magdeburg;

			3Institute of Biology, Otto von Guericke University Magdeburg; 4Center for Behavioral Brain Sciences, Magdeburg
Akinao Nose	Embryonic development of the motor circuits in Drosophila: emergence of coordinated neural activities and the role of sensory feedback	Xiangsunze Zeng[1], Tappei Kawasaki[1], Kengo Inada[2], Hokto Kazama[2,4], Akinao Nose[1,3] 1. Dept. of Comp. Sci. Eng., Grad. Sch. of Frontiers Science, The Univ. of Tokyo 2. RIKEN Center for Brain Science 3. Dept. of Physics, Grad. Sch. of Science, The Univ. of Tokyo 4. Dept. of Life Sci., Grad. Sch. Arts & Sci, The Univ. of Tokyo	Animals' motor patterns form in a gradual manner during late embryogenesis as the innervation of the somatic musculature and connectivity within the central nervous system develop. Initial uncoordinated or premature motor activity emerges while the animals are still in the womb or egg and reflects the initiation of functional locomotor circuits. For instance, in the spinal cord of vertebrates, initial bursts occur in local groups of neurons and induce contractions of the target muscles. It has been proposed that such premature motor activities, via sensory feedback of the muscular movements, instruct the formation of functional locomotor circuits. However, little is known about the underlying circuit mechanism and molecular basis. In this study, we used peristaltic locomotion of larval Drosophila to investigate the role of proprioceptive experience in formation of locomotor circuits. We carried out calcium imaging and patch-clamp recordings of the isolated central nervous system to study the development of the central pattern generators (CPGs) that drive peristaltic locomotion. Here, we found that in nompC and tmc mutants, which lack sensory feedback of muscular movement, the CPGs fail to develop properly. Our results of dye-coupling also suggest that gap-junctional transmission in a target interneuron of the proprioceptor is required for proper function of the CPGs. Based on these and other results, we will discuss how sensory feedback might regulate the development of the functional locomotor circuits from molecular to behavioral perspectives.
Xiangsunze Zeng	Embryonic development of the motor circuits in *Drosophila*: emergence of coordinated neural activities and the role of sensory feedback	Xiangsunze Zeng[1], Tappei Kawasaki[1], Kengo Inada[2], Hokto Kazama[2,4], Akinao Nose[1,3] 5. Dept. of Comp. Sci. Eng., Grad. Sch. of Frontiers Science, The Univ. of Tokyo 6. RIKEN Center for Brain Science 7. Dept. of Physics, Grad. Sch. of Science, The Univ.	Animals' motor patterns form in a gradual manner during late embryogenesis as the innervation of the somatic musculature and connectivity within the central nervous system develop. Initial uncoordinated or premature motor activity emerges while the animals are still in the womb or egg and reflects the initiation of

		of Tokyo 8. Dept. of Life Sci., Grad. Sch. Arts & Sci, The Univ. of Tokyo	functional locomotor circuits. For instance, in the spinal cord of vertebrates, initial bursts occur in local groups of neurons and induce contractions of the target muscles. It has been proposed that such premature motor activities, via sensory feedback of the muscular movements, instruct the formation of functional locomotor circuits. However, little is known about the underlying circuit mechanism and molecular basis. In this study, we used peristaltic locomotion of larval *Drosophila* to investigate the role of proprioceptive experience in formation of locomotor circuits. We carried out calcium imaging and patch-clamp recordings of the isolated central nervous system to study the development of the central pattern generators (CPGs) that drive peristaltic locomotion. Here, we found that in *nompC* and *tmc* mutants, which lack sensory feedback of muscular movement, the CPGs fail to develop properly. Our results of dye-coupling also suggest that gap-junctional transmission in a target interneuron of the proprioceptor is required for proper function of the CPGs. Based on these and other